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Diadema ahmadii (*Pleosporales*), a new ascomycetous species from Pakistan

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Abstract — *Diadema ahmadii* sp. nov. is described, illustrated, and compared with similar taxa. This species was collected from dead branches of *Rosa moschata* in Kaghan Valley, an alpine region in Pakistan. *Diadema ahmadii* is most similar to *D. tetramerum*, the type species of the genus, in that it has asci and ascospores of similar dimensions. However, *D. ahmadii* is distinguished from the latter and other related species by having ascospores with a submedian primary septum.

Key words — bitunicate ascomycetes, *Diademaceae*, *Dothideomycetes*, *Pleosporomycetidae*

Introduction

During the examination of several herbarium specimens of bitunicate ascomycetes in Pakistan, an interesting species with dark and relatively large ascospores was found on dead branches of *Rosa moschata* collected from an alpine region in Pakistan (Batakundi, Kaghan Valley). Owing to the presence of globose to subglobose ascomata without a papillate beak, obclavate to cylindrical asci with fissitunicate dehiscence, and deeply pigmented, 3-septate ascospores, this ascomycete was considered as an undescribed species in the genus *Diadema* Shoemaker & C.E. Babc. The new species is described, illustrated, and compared to other species in this genus.

Materials and methods

Microscopical observations followed methods as described in Tanaka & Harada (2003) and Tanaka et al. (2009) have been followed. To observe the internal structure of ascospores, 5% NaCLO was used to bleach strongly melanized ascospores as described in Eriksson (1989). Ratios indicating ascospore septum

position follow Shoemaker (1984; length of upper hemispore/total length of ascospore). Holotype and isotype specimens were deposited in the herbaria of LAH (SHI Mycological Herbarium, University of the Punjab) and HHUF (Hirosaki University), respectively.

Taxonomy

Diadema ahmadii Kaz. Tanaka & S.H. Iqbal, **sp. nov.** Figs. 1–15

MycoBank MB 513556

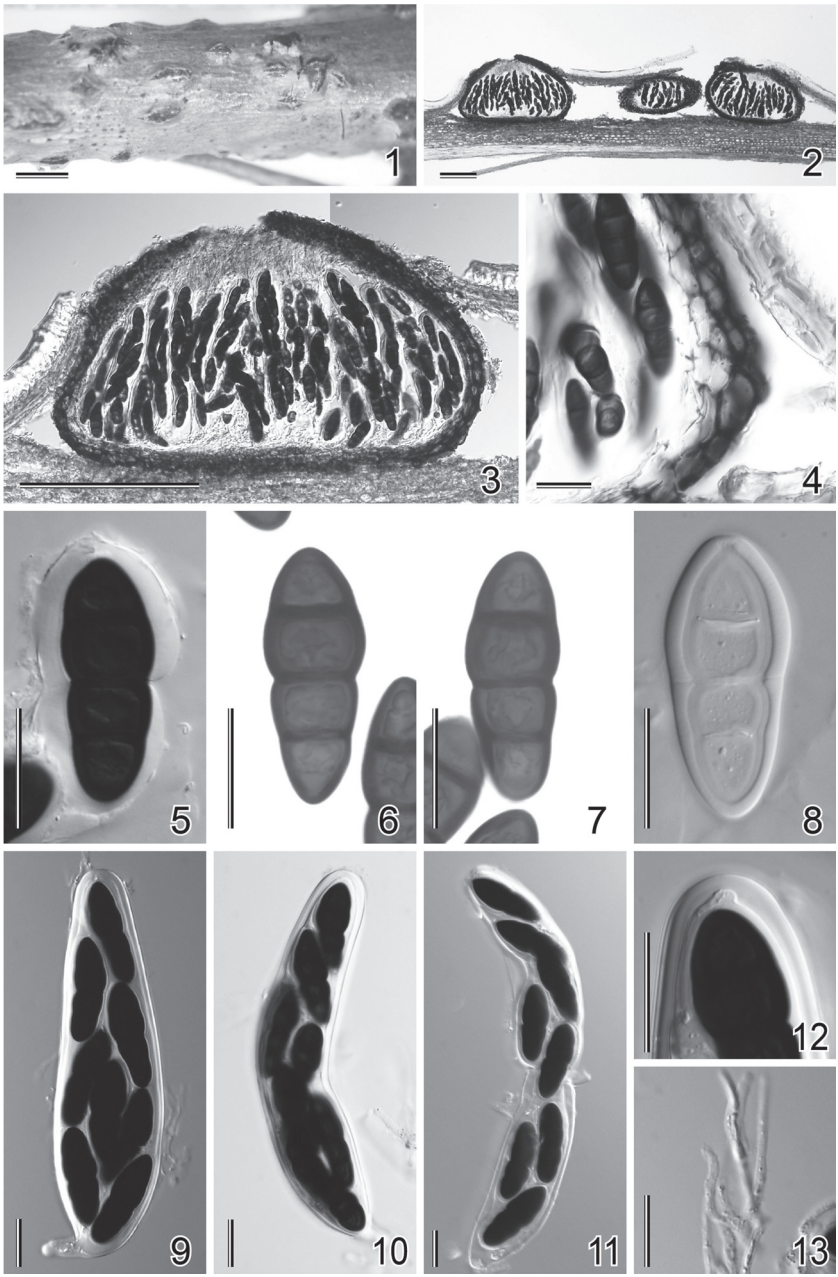
Ascomata 240–310 µm alta, 290–500 µm diametro, immersa, erumpentia ad apicem, dispersa vel 2–4 congregata, unilocularia, globosa vel subglobosa, glabrata. Orificinum centrale, planum, non papillatum. Paries ascomatis uniformiter 12–30 crassus. Pseudoparaphyses 2–4 µm latae. Asci (135–)140–185(–193) × 29–42(–44.5) µm, obclavati vel cylindrici, apice rotundati, stipitati, fissitunicati, octospori. Ascosporae 37–44(–47) × 13–16 µm, cum septo primum submedio (0.53), 3-septatae, late fusiformis, crassitunicatae, badiae vel fere nigerae, strato mucoso 2–4 µm lato circumdatae.

Holotype: **PAKISTAN**. Northwestern Frontier Province, Lalazar (Batakundi), Kaghan Valley, on dead branches of *Rosa moschata*, 3 September 1967, SHI2007-541, LAH (**holotype**), HHUF 30004 (**isotype**).

Etymology: named in honor of Dr. Sultan Ahmad for his outstanding work on Pakistan fungi.

Ascomata 240–310 µm high, 290–500 µm diam, immersed, erumpent at the apex, scattered to 2–4 gregarious, uniloculate, globose to subglobose with flattened base in longitudinal section, glabrous. Opening area central, flat, not papillate; lid not seen. Ascomatal wall of 'textura angularis' in surface view; wall in longitudinal section uniformly 12–30 µm thick at sides, composed of 4–6 layers of polygonal cells (12–25 × 4–13 µm); wall at base composed of rectangular to subglobose cells of 5–12 µm diam; wall around the opening area composed of flattened dark brown cells of 5–8 × 2–5 µm. Pseudoparaphyses numerous, sometimes very sparse, septate, branched and anastomosed, 2–4 µm wide. Asci (135–)140–185(–193) × 29–42(–44.5) µm (mean = 159.3 × 36.6 µm, n = 50), obclavate to cylindrical, rounded at the apex, with a shallow ocular chamber, broadly at below, stipitate, fissitunicate, with 8 biseriate ascospores. Ascospores 37–44(–47) × 13–16 µm (mean = 41.3 × 15.0 µm, n = 70), L/W 2.5–3.1 (mean = 2.8, n = 70), with a primary septum submedian (0.51–0.55; mean = 0.53, n = 70), 3-septate, rarely with an additional septum at the basal cell, broadly fusiform, mostly straight, enlarged at second cell from apex, strongly constricted at primary septum, weakly constricted at other septa,

Figs. 1–13. *Diadema ahmadii*. 1. Ascomata on the host surface. 2–3. Longitudinal section of ascomata. 4. Ascomatal wall in longitudinal section. 5–7. Ascospores in water mount. 8. Ascospore in NaClO, 9–10. Asci. 11. Extending fissitunicate ascus. 12. Apex of ascus. 13. Pseudoparaphyses. Bars: 1 = 1 mm, 2–3 = 200 µm, 4–13 = 20 µm.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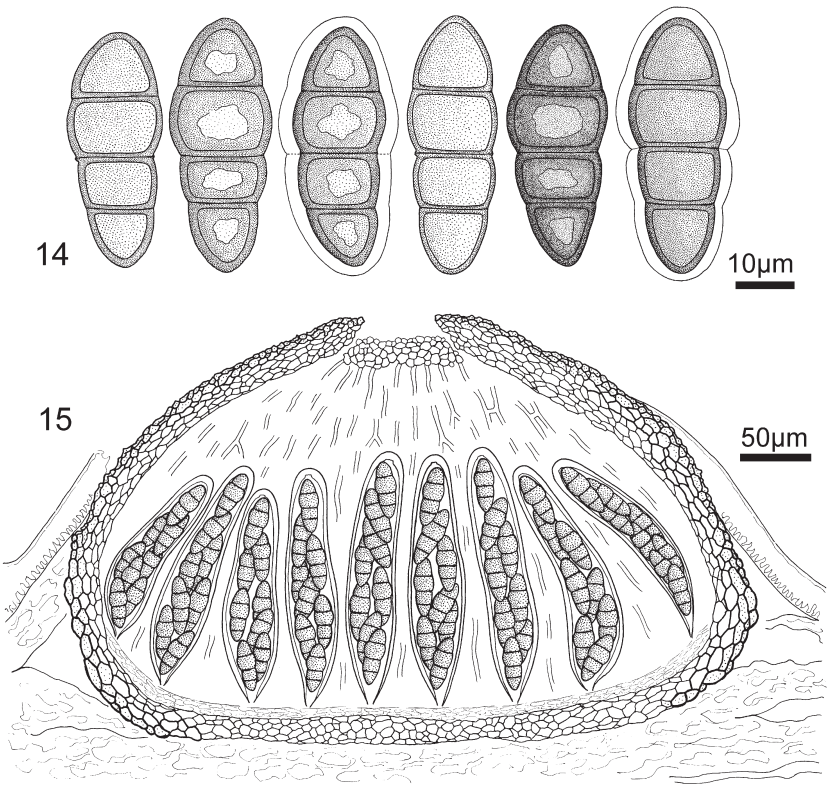

Figs. 14–15. Line drawings of *Diadema ahmadii*.
14. Ascospores. 15. Ascoma in longitudinal section. Bars: 14 = 10 µm. 15 = 50 µm.

thick-walled (ca. 1–2 µm wide), reddish brown to almost black, smooth, with a sheath. Sheath entire, firm, sharply delimited, 2–4 µm thick, mostly constricted at the side of primary septum.

Discussion

Diadema, an ascomycetous genus typified by *D. tetramerum* Shoemaker & C.E. Babc., is assigned to *Diademaceae*, a family characterized by the presence of a 'lid' or 'cap' in the area of the ascomatal opening (Shoemaker & Babcock 1992). Most members of *Diadema* are known from culms or stems of alpine plants, in particular, those belonging to *Poaceae* or *Rosaceae*, and are reported from India, Pakistan, and USA (Shoemaker & Babcock 1989). The characteristic features of *Diadema* are relatively large, deeply pigmented ascospores and the disc-like

opening system of the ascomata. These morphological ascomycete features are generally regarded as adaptations to severe alpine conditions, such as high UV-radiation and low temperature (Savile 1972, Leuchtmann 1987, Shoemaker & Babcock 1989). *Diadema* was monographed by Shoemaker & Babcock (1989), who accepted 6 species in this genus. Subsequently, one species was added by Huhndorf (1992).

The overall morphological features of *Diadema ahmadii* as well as its alpine habitat in Pakistan agree with the current concept of *Diadema*. However, we could not find a 'lid' or 'cap' at the opening of the ascomatal apex in our material. This cap-like structure is also absent in other species of *Diadema*, such as *D. sieversiae* (Peck) Huhndorf and *D. obtusum* Shoemaker & C.E. Babc. (Shoemaker & Babcock 1987, Huhndorf 1992). The opening area of the ascomata in the Pakistan material was obscure, but the presence of a flattened apex in ascomata that lacked a papillate beak, the wall around the opening area composed of small flattened dark brown cells, and the subtending pseudoparaphyses suggest that the opening system of *D. ahmadii* might be essentially the same as that of other species in *Diadema*.

Table 1. Comparison of *Diadema ahmadii* with other species in the genus *Diadema*

Taxa	Ascospores				Hosts	Distribution
	Size (µm)	L/W	PS*	Septa		
D. ahmadii (this study)	37–44(–47) × 13–16	2.8	0.53	3	*Rosaceae*	Pakistan
D. acutum Shoemaker & C.E. Babc. [1)]	50–56 × 16–21	2.9	0.50	3	*Poaceae*	India, USA
D. cinctum Shoemaker & C.E. Babc. [1)]	33–38 × 13–18	2.3	0.50	3	*Cyperaceae*	India
D. curtum Shoemaker & C.E. Babc. [1)]	43–49 × 15–20	2.7	0.50	3	*Biebersteiniaceae*	India
D. hexamerum Shoemaker & C.E. Babc. [1)]	40–50 × 15–17	2.6	0.50	5	*Poaceae*	USA
D. obtusum [1)]	(40–)46–50(–55) × 19–21	2.3	0.50	3	*Rosaceae*, *Poaceae*	India, Pakistan
D. sieversiae [2) 3)]	50–56 × 20–28	–	0.48	3	*Rosaceae*, *Ericaceae*	USA
D. tetramerum [1)]	36–50 × 14–20	2.7	0.50	3	*Juncaceae*, *Poaceae*	USA

Data from [1)] Shoemaker & Babcock (1989), [2)] Shoemaker & Babcock (1987), [3)] Huhndorf (1992).
* PS = position of the primary septum (length of upper hemispore/total length of ascospore).

Among the 7 species previously recognized in this genus, *D. ahmadii* superficially resembles *D. tetramerum*, the type species of the genus, in having asci and ascospores of similar dimensions. Like *D. ahmadii*, the species, *D. obtusum* and *D. sieversiae*, have also been recorded on host plants belonging to the family *Rosaceae* in alpine regions. *Diadema ahmadii*, however, can be easily distinguished from all known *Diadema* species owing to the presence of ascospores with submedian primary septum. A synopsis of these differences is shown in Table 1.

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